

DNA Isolation from Whole Blood

DNA extraction from liquid blood using *forensicGEM*[™]

The following method is recommended for DNA extraction from human blood samples using *forensicGEM*[™]. This method is optimised for fresh blood or blood stabilised with Heparin, Citrate or EDTA.

Extraction Method

1. Add 2.5 µl of blood to 96.5 µl of *forensicGEM*[™] buffer 3H and 1 µl of *forensicGEM*[™] [IMPORTANT - NOTE1; EDTA blood].
2. Incubate at 75 °C for 15 minutes.
3. Incubate at 95°C for 5 - 15 minutes [NOTE2].
4. Centrifuge the extract at full speed for 5 minutes to separate the phases.
5. Typically, 0.5 – 1 µl of the supernatant should be used for profiling [NOTE3].

NOTE1: *forensicGEM*[™] is Ca²⁺ dependent, so for EDTA stabilised blood, the buffer should be supplemented to a final concentration of 200 µM CaCl₂. This concentration will be effective for Vacutainers containing between 1 and 7 ml. *forensicGEM*[™] and buffer can also be added as a master mix.

NOTE2: 5 minutes is sufficient for fresh blood and blood stabilised with Heparin or Citrate. *forensicGEM*[™] is stabilised by Ca²⁺ and so for EDTA stored blood where extra calcium has been added, a 15 minute heat-kill is required.

NOTE3: This procedure yields on average 6 ng/µl. Quantiblot tends to underestimate quantity of amplifiable DNA and is only a guide. Use a factor of 20 x when using Pico Green.

WHOLE BLOOD

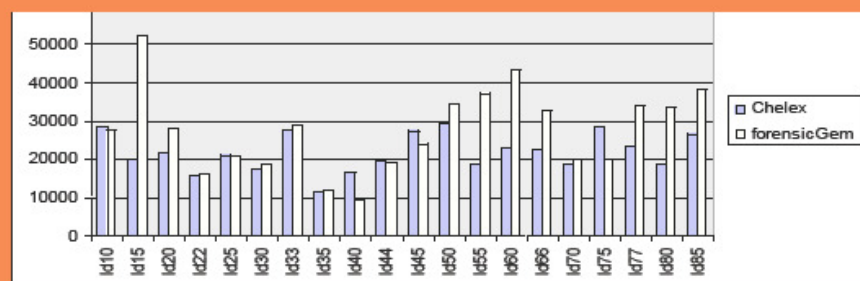
The results below were from an extraction from 2.5 µl of fresh blood. 1 µl of the extract was profiled using AmpFlSTR[®] Identifiler[™] PCR Amplification Kit (Applied Biosystems).



Top Panel: *forensicGEM*[™] extracted DNA.

Middle Panel: Manufacturer supplied control DNA.

Bottom Panel: No *forensicGEM*[™] added to extraction



The histogram compares the total profile peak heights obtained from 20 replicates using the *forensicGEM*[™] method and a standard method using Chelex[®].